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Enhancing effect of dipyridamole inhalation on adenosine-induced bronchospasm in asthmatic patients

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The study was performed on 13 asthmatic patients to determine whether inhaled dipyridamole would act directly by inducing bronchoconstriction or indirectly by potentiating the adenosine-induced bronchoconstriction. The study was performed in 3 consecutive days. On the first day adenosine challenge was performed and the PD_{20} value calculated. On the other days the adenosine challenge was done 5 min after randomized inhalations of dipyridamole or a control solution. The mean percent change in FEV_1 after dipyridamole (Δ % = 2.0) and control solution (Δ % = 1.0) was not significant. Inhaled adenosine caused bronchoconstriction with a geometric mean PD_{20} of 1.09 mg. After control solution inhalation, a mean PD_{20} value of 1.31 mg was observed. Dipyridamole inhalation increased adenosine hyperresponsiveness and in all subjects shifted the dose-response curves of adenosine challenge to the left with a mean PD_{20} value of 0.40 mg. This enhancing effect of dipyridamole was significant when compared with the baseline value ($P < 0.01$) and control solution ($P < 0.01$). The study demonstrated that dipyridamole inhalation increased airway responsiveness to adenosine in all subjects. This effect is due to indirect activity of dipyridamole on airways without changes in baseline airway caliber.

Key words: adenosine; adenosine-induced bronchospasm; airway hyperresponsiveness; dipyridamole.

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Coleman & Levy (4) demonstrated the spasmolytic activity of adenosine and adenosine 5'-triphosphate (ATP) in a guinea pig isolated tracheal preparation.

This spasmolytic activity was enhanced by dipyridamole which was reported to inhibit adenosine uptake (16). This inhibition in the uptake of adenosine leads to the accumulation of extracellular adenosine and thereby causes an increase in blood flow (18).

Several studies with isolated tracheal preparations have shown that adenosine produes a relaxation (1, 2, 5, 6, 13, 15). This response has been attributed to an increase in intracellular cAMP through stimulation of adenylate cyclase via purine receptors of the PI type (3). Adenosine affects adenylate cyclase via two types of extracellular receptors: one a high affinity inhibitory receptor (A_1), and the other (A_2) a low affinity stimulatory receptor (12, 20).

It would be expected that adenosine acts as a bronchodilating agent by stimulating airway smooth muscle adenylate cyclase in the same way as β_2-adrenergic drugs. However, previous studies (8,9,10,17) and our previously reported observations (7) demonstrated that adenosine-induced bronchoconstriction and this mechanism is unknown.

Dipyridamole is a potent inhibitor of adenosine uptake; but no significant bronchodilating effect has been previously demonstrated by oral dipyridamole and it did not enhance the effect of β_2-adrenergic drugs (19).

In a previous study Cushley et al. (11)

demonstrated that intravenous infusion of dipyridamole had a small non-significant enhancing effect on adenosine-induced bronchoconstriction in asthmatic subjects. The effect of dipyridamole by inhalation was not investigated.

The aim of this study was to determine whether inhaled dipyridamole would act directly by inducing bronchoconstriction or act to potentiate the adenosine-induced bronchoconstriction in asthmatic subjects.

PATIENTS AND METHODS

Subjects

The study was performed on 13 asthmatic patients (3 male and 9 female, age range 17 to 46 years). All were selected from the Allergy Unit of Respiratory Disease in Catania. All had a history of dyspnea with wheezing or chest tightness on exposure to one or more airborne allergens and all had a positive skin test (Prick) to one or more common allergens. They were non-smokers with normal chest X-rays and none had features of other respiratory disease. At the start of the study all patients were asymptomatic with FEV_1 values not lower than 70% of their predicted normal value (Table 1). The PD_{20} value of adenosine challenge was measured prior to accepting the patients to the clinical study. All therapeutic treatment was discontinued at least 48 h before the study. During the trial, patients avoided taking food or drinks (tea, chocolate, coffee) which could interfere with the study.

The protocol was approved by the Ethics Review Committee and consent was obtained from each subject after the nature and reason for the study had been explained in detail.

Study design

The study was conducted in 3 consecutive days at the same time each morning. On the first day adenosine challenge was performed and the PD_{20} value calculated. On the following 2 days the adenosine challenge was done 5 min after randomized inhalations of dipyridamole or control solution.

Inhalation test

The inhalation solutions were administered as aerosols generated by a nebulizer DeVilbiss 646 with 2 ml solution and an airflow of 12 l/min. Adenosine (Merck 862/1451739) was prepared in 0.9% saline to produce a dose range 0.03–4 mg. Adenosine challenge was performed by the standardized technique previously described (7). A dose (2.5 mg) of dipyridamole solution (Boeringher Ingelheim) was administered by 15 tidal breathings.

The dipyridamole comprised:

– dipyridamole	2.5	mg
– distilled water	0.946	ml
– polyethylene glycol 600	50	mg
– HCl 1N	9	mg
– tartaric acid	2	mg

The solution in which dipyridamole was diluted was used as control. Dipyridamole and control solution were administered 5 min before the adenosine challenge, and FEV_1 was initially measured at 1, 3 and 5 min intervals and then every 5 min until a higher value than the previous was obtained. The dose of adenosine producing 20% change in FEV_1 was calculated from the individual semilogarithmic dose-response curve (PD_{20}).

Statistical analysis

Mean PD_{20} and Δ % values were calculated using logarithmic transformation.

Statistical analysis was performed by Student's paired t-test in order to compare changes in bronchial hyperreactivity and FEV_1. A Wilcoxon's signed rank test was performed to compare the percentage of variation.

RESULTS

Baseline FEV_1 did not differ significantly on the 3 study days with mean values of 2.84 ± 0.27 l, 2.92 ± 0.37 l and 2.86 ± 0.32 l on the first day, the dipyridamole and the control days, respec-

Table 1

Patient characteristics

Patient No.	Sex	Age (years)	Height (cm)	Weight (kg)	Predicted FEV_1 (l)	Measured FEV_1 (l)	% Pred.	PD_{20} adenosine (mg)
1	M	40	159	73	3.26	2.70	82	1.90
2	M	25	174	76	4.16	3.43	82	0.74
3	F	29	157	51	3.00	2.87	95	1.40
4	F	34	153	64	2.81	2.73	97	1.60
5	F	46	150	64	2.49	2.76	110	0.95
6	F	28	168	74	3.29	2.63	80	0.68
7	F	17	160	55	2.75	3.14	114	0.91
8	F	32	154	64	2.87	2.73	94	1.25
9	F	38	152	65	2.70	2.36	87	1.70
10	M	46	176	78	3.73	2.94	78	1.05
11	F	33	160	66	2.99	2.83	95	0.70
12	F	29	160	56	2.94	3.10	102	1.02
M		33.08	160.2	65.5	3.08	2.84	93	1.09*
SD		± 8.48	± 8.4	± 8.6	± 0.467	± 0.27	± 11.71	1.55**

*Geometric mean
**Geometric mean + 1 SD

Table 2

Mean (± SD) Baseline 1 FEV_1 and after dipyridamole and control solution (Baseline 2)

Treatment	Baseline 1	Baseline 2	%
Dipyridamole	2.92 ± 0.37	2.86 ± 0.40	2.0
Control solution	2.86 ± 0.32	2.83 ± 0.36	1.0

Table 3

Effect of dipyridamole and control solution on PD_{20} adenosine

Patient No.	Baseline	Dipyridamole	Control solution
1	1.90	0.60	2.60
2	0.74	0.32	1.56
3	1.40	0.25	1.88
4	1.60	0.57	2.24
5	0.95	0.37	1.12
6	0.68	0.25	0.49
7	0.91	0.27	0.37
8	1.25	0.79	2.70
9	1.70	0.55	2.20
10	1.05	0.29	1.38
11	0.70	0.25	0.43
12	1.02	0.70	2.10
Geometric mean	1.09	0.40*	1.31
G.M. + SD	1.55	0.62	2.70

*$P < 0.01$ vs baseline and control

tively (Table 2). A small effect on baseline airway caliber was noticed after inhalation of dipyridamole and control solution, with mean values of 2.86 ± 0.40 and 2.83 ± 0.36, respectively. The mean percent change in FEV_1 after dipyridamole (Δ % = 2.0) and control solution (Δ % = 1.0) was not significant.

Inhaled adenosine caused bronchoconstriction with a geometric mean PD_{20} of 1.09 mg. After control solution inhalation, a mean PD_{20} of 1.31 mg was observed which was not significantly different from the mean baseline value (Table 3). Dipyridamole inhalation increased adenosine hyperresponsiveness and in all subjects shifted the dose-response curves of adenosine challenge to the left with a mean PD_{20} value of 0.40 mg (Table 3). This enhancing effect of dipyridamole was significantly different when compared with baseline value ($P < 0.01$) and control solution ($P < 0.01$). The mean percent change (Δ % = −14.5) in PD_{20} value (Fig. 1) after dipyridamole inhalation was significant ($P < 0.01$) compared with that obtained after control solution (Δ % = +2.5).

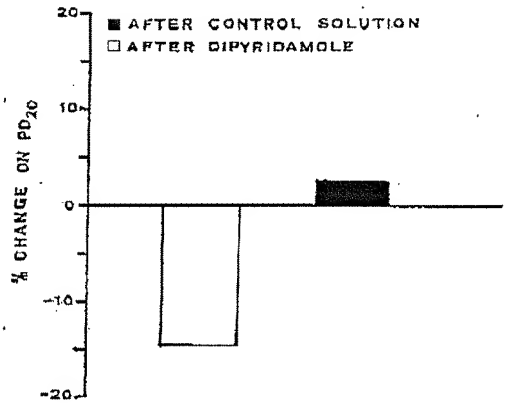

Fig. 1. Percent changes in PD_{20} after inhalation of dipyridamole and control solution.

DISCUSSION

The study confirms previous findings that adenosine induced bronchoconstriction in asthmatic patients (7, 9, 10).

Dipyridamole inhalation increased airway responsiveness to adenosine in all subjects. This enhancement is evident with dipyridamole but not with control solution. This effect is due to the activity of dipyridamole on airways without changes in baseline airway caliber. Adenosine was more potent in the presence of dipyridamole, thus confirming previous *in vitro* studies (5, 13, 14).

Dipyridamole is known to inhibit the uptake of adenosine, and the enhancement of adenosine activity on airways can be attributed to this property. This suggested that the bronchoconstriction induced by adenosine is mediated via extracellular adenosine receptors located on the cell membrane. Cushley et al. (11) demonstrated that infused dipyridamole was not able to enhance significantly airway reactivity to adenosine even if a small decrease in the concentration of adenosine was noticed in the presence of dipyridamole.

Our results suggest that dipyridamole influences directly the uptake of adenosine in airway tissue and probably acts directly on surface adenosine receptors, reaching a higher concentration in bronchial tissue by inhalation rather than infusion.

Infused dipyridamole could inhibit adenosine uptake to a higher extent in circulating platelets than in cell tissue, probably because adenosine was rapidly inactivated.

The mechanism of adenosine is still unknown even if several studies (12, 20) have demonstrated a direct action of adenosine on specific receptors.

In conclusion, our study demonstrated the potentiating effect of dipyridamole by inhalation on adenosine broncoconstriction. However, further studies are necessary to clarify the exact mechanism of adenosine bronchoconstriction in asthmatic subjects.

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